

AMENDMENTS TO THE CLAIMS

This listing of claims replaces all prior versions of claims in the application.

1. (Currently Amended) A method for quantifying [[a]] small particle low density lipoprotein LDL in a test sample, comprising:

(i) removing lipoproteins other than a first step for separating the small particle low density lipoprotein from other low density lipoproteins, and a second step for measuring cholesterol, triglycerides or proteins in the separated small particle low density lipoprotein LDL and HDL from said test sample; and then

(ii) quantifying small particle LDL in said test sample from step (i) by measuring the amount of LDL,

wherein step (i) comprises adding a separation agent comprising a polyanion and a divalent cation to said test sample.

2. (Cancelled)

3. (Currently Amended) A method according to claim 1 [[or 2]], wherein said separation agent further comprises a monovalent cation is further used for separating the small particle low density lipoprotein from other low density lipoproteins in said first step.

4. (Currently Amended) A method according to claim 12 or 3, wherein the polyanion used in said first step is selected from the group consisting of heparin, phosphotungstic acid and dextran sulfate.

5. (Currently Amended) A method according to any one of claims 2 to 4 claim 1, wherein the divalent cation used in said first step is selected from the group consisting of Mn²⁺, Mg²⁺ and Ca²⁺.

6. (Currently Amended) A method according to any one of claims 3 to 5 claim 3, wherein the monovalent cation used in said first step is selected from the group consisting of Na⁺, K⁺ and Li⁺.

7. (Currently Amended) A method according to any one of claims 4 to 6 claim 4, wherein, when the polyanion is added to the test sample, the final concentration of the polyanion

is 10-250 U/mL for heparin, 0.02-1.25% for dextran sulfate and 0.02-1.25% for phosphotungstic acid.

8. (Currently Amended) A method according to ~~any one of claims 5 to 7~~ claim 5, wherein, when the divalent cation is added to the test sample, the final concentration of the divalent cation is 2.5-35 mmol/L for Mn²⁺, 2.5-125 mmol/L for Mg²⁺ and 1-75 mmol/L for Ca²⁺.

9. (Currently Amended) A method according to ~~any one of claims 6 to 8~~ claim 6, wherein, when the monovalent cation is added to the test sample, the final concentration of the monovalent cation is 0-50 mmol/L.

10. (Currently Amended) A method ~~according to claim 1, for quantifying small particle LDL in a test sample, comprising:~~

~~(i) removing lipoproteins other than small particle LDL and HDL from said test sample; and then~~

~~(ii) quantifying small particle LDL in said test sample from step (i) by measuring the amount of LDL,~~

~~wherein step (i) comprises adding PEG to said test sample, is used to separate the small particle low density lipoprotein from other low density lipoproteins in said first step.~~

11. (Currently Amended) A method according to claim 10 wherein the final concentration of PEG is 2-5% by weight when PEG is added to the test sample.

12. (Currently Amended) A method according to ~~any one of claims 1 to 11~~ claim 1, wherein ~~measuring the amount of LDL the measurement of cholesterol in said second step is carried out by using a reagent which is used for quantitatively selectively measuring cholesterol in a low density lipoprotein LDL and which does not require fractionation.~~

13. (Currently Amended) A method according to ~~any one of claims 1 to 11~~ claim 1, wherein ~~measuring the amount of LDL the measurement of triglycerides in said second step is carried out by using a reagent which is used for quantitatively selectively measuring triglycerides in a low density lipoprotein LDL and which does not require fractionation.~~

14. (Currently Amended) A method according to ~~any one of claims 1 to 11~~ claim 1, wherein ~~measuring the amount of LDL the measurement of protein in said second step~~ is carried out by using an anti-human apoprotein B antibody.

15. (Currently Amended) A method for separating [[a]] small particle ~~low density lipoprotein LDL~~ from a test sample ~~that contains LDLs~~, comprising a step in which the low density lipoprotein ~~precipitating LDLs~~ other than small particle low density lipoproteins is precipitated ~~LDL~~ by adding a separation agent comprising a polyanion and a divalent cation to the test sample.

16. (Currently Amended) A method according to claim 15, ~~wherein said separation agent further comprises a monovalent cation comprising a step in which the low density lipoprotein other than small particle low density lipoproteins is precipitated by also adding a monovalent cation to the test sample.~~

17. (Currently Amended) A method for separating a small particle ~~low density lipoprotein~~ according to claim 15 [[or 16]], wherein the polyanion is selected from the group consisting of heparin, phosphotungstic acid and dextran sulfate.

18. (Currently Amended) A method for separating a small particle ~~low density lipoprotein~~ according to ~~any one of claims 15 to 17~~ claim 15, wherein the divalent cation is selected from the group consisting of Mn²⁺, Mg²⁺ and Ca²⁺.

19. (Currently Amended) A method for separating a small particle ~~low density lipoprotein~~ according to ~~any one of claims 15 to 18~~ claim 15, wherein the monovalent cation is selected from the group consisting of Na⁺, K⁺ and Li⁺.

20. (Currently Amended) A method for separating a small particle ~~low density lipoprotein~~ according to ~~any one of claims 17 to 19~~ claim 17, wherein, when the polyanion is added to the test sample, the final concentration of the polyanion is 10-250 U/mL for heparin, 0.02-1.25% for dextran sulfate and 0.02-1.25% for phosphotungstic acid.

21. (Currently Amended) A method for separating a small particle ~~low density lipoprotein~~ according to ~~any one of claims 18 to 20~~ claim 18, wherein, when the divalent cation

is added to the test sample, the final concentration of the divalent cation is 2.5-35 mmol/L for Mn²⁺, 2.5-125 mmol/L for Mg²⁺ and 1-75 mmol/L for Ca²⁺.

22. (Currently Amended) A method ~~for separating a small particle low density lipoprotein according to any one of claims 19 to 21~~ claim 16, wherein, when the monovalent cation is added to the test sample, the final concentration of the monovalent cation is 0-50 mmol/L.

23. (Currently Amended) A method for separating a small particle low density lipoprotein from a test sample ~~that contains LDLs~~, comprising a step in which PEG is added to the test sample to precipitate the low density lipoprotein precipitating LDLs other than small particle low density lipoproteins LDL by adding PEG to the test sample.

24. (Currently Amended) A method ~~for separating a small particle low density lipoprotein according to claim 23~~, wherein the final concentration of PEG is 2-5% by weight when PEG is added to the test sample.

25. (Currently Amended) A kit for measuring [[a]] small particle ~~low density lipoprotein LDL in a test sample~~, comprising:

(i) a surface active agent, which is selected from the group consisting of polyoxyethylene lauryl ether, polyoxyethylene cetyl ether, polyoxyethylene octylphenyl ether and polyoxyethylene nonylphenyl ether;

(ii) a separation agent that includes comprises a polyanion and a divalent cation, wherein the polyanion is selected from the group consisting of heparin, phosphotungstic acid and destran sulfate; and

(iii) a reagent for measuring the low density lipoprotein LDL via measuring, wherein the kit measures cholesterol, triglycerides or proteins in the small particle low density lipoprotein LDL.

26. (Currently Amended) A kit ~~for measuring a small particle low density lipoprotein~~ according to claim 25, wherein the separation agent further includes comprises a monovalent cation.

27. (Currently Amended) A kit for measuring [[a]] small particle low density lipoprotein LDL in a test sample, comprising:

- (i) a surface active agent, which is selected from the group consisting of polyoxyethylene lauryl ether, polyoxyethylene cetyl ether, polyoxyethylene octylphenyl ether and polyoxyethylene nonylphenyl ether;
- (ii) a separation agent that includes comprises PEG; and
- (iii) a reagent for measuring the low density lipoprotein LDL via measuring, wherein the kit measures cholesterol, triglycerides or proteins in the small particle low density lipoprotein LDL.

28. (Cancelled)

29. (Currently Amended) A kit according to claim 26 or 28, wherein the divalent cation is selected from the group consisting of Mn²⁺, Mg²⁺ and Ca²⁺ and the monovalent cation is selected from the group consisting of Na⁺, K⁺ and Li⁺.

30. (New) A kit according to claim 25, wherein the separation agent comprises 60 U/mL of sodium heparin and 40 mmol/L of MnCl₂, and wherein the separation agent is added to the test sample at a volume ratio of 1:1.

31. (New) A kit according to claim 25, wherein the separation agent comprises 300 U/mL of sodium heparin and 150 mmol/L of MgCl₂, and wherein the separation agent is added to the test sample at a volume ratio of 1:1.

32. (New) A kit according to claim 25, wherein the separation agent comprises 1.5% dextran sulfate with an average molecular weight of 5000 and 40 mmol/L of MgCl₂, and wherein the separation agent is added to the test sample at a volume ratio of 1:1.

33. (New) A kit according to claim 25, wherein the separation agent comprises 0.3% sodium phosphotungstic acid and 7.5 mmol/L of CaCl₂, and wherein the separation agent is added to the test sample at a volume ratio of 1:1.

34. (New) A kit according to claim 25, wherein the separation agent comprises 40 U/mL sodium heparin and 30 mmol/L of MnCl₂, and wherein the separation agent is added to the test sample at a volume ratio of 1:1.

35. (New) A kit according to claim 25, wherein the separation agent comprises 500 U/mL sodium heparin, 140 mmol/mL MgCl₂ and 34 mmol/L of KCl, and wherein the separation agent is added to the test sample at a volume ratio of 1:1.

36. (New) A kit according to claim 25, wherein the separation agent comprises 150 U/mL sodium heparin, 90 mmol/mL MgCl₂, and wherein the separation agent is added to the test sample at a volume ratio of 1:1.